Rasayely
International Journal of Equine Science
https://rasayely-journals.com/index.php/ijes
Vol 1(1); 16–32, 2022

Review Article Open Access

Penetration Enhancers for the Development of Intranasal Formulations for Use in Equines

María Inés Velloso[1] and Fabiana Landoni[1,*]

[1]*Faculty of Veterinary Sciences, National University of La Plata, La Plata, Buenos Aires, Argentina*

[*]Author to whom any correspondence should be addressed; e-mail: landoni@fcv.unlp.edu.ar, Tel. +54-0221-423-6663/64424-9621

Received: 14 October 2021; Revised: 11 January 2022; Accepted: 14 March 2022; Published: 28 March 2022

Academic Editor: Guilherme Camargo Ferraz, São Paulo State University, Brazil

Abstract

The aim of this review is to assess penetration enhancers, such as cyclodextrins, chitosan and their derivatives, surfactants, bile acids, their salts and derivatives, sodium taurodihidrydrofusidate, and phospholipids used in the development of intranasal formulations with a potential application in horses. In the last few years, the interest in the intranasal administration route in humans has grown because it is bloodless, noninvasive, and painless and represents a direct path toward the central nervous system. However, in equine medicine, the use of this administration route is scarce. Since equines have a nasal cavity with large surface area and blood irrigation, a high bioavailability of intranasal administered drugs is expected. Nowadays, the development of formulations for intranasal administration in equines is a challenge. The present review proposes the assessment of the characteristics and potential effects of the most important penetration enhancers in the development of intranasal formulations for use in equines.

Keywords

Horses; cyclodextrins; chitosan; surfactants; bile acids; sodium taurodihidrydrofusidate; phospholipids

1. Introduction

Nowadays, the development of formulations for intranasal administration in equines is a challenge. When a drug is administered intranasally, before arriving at the action site, it must face many obstacles that are peculiar to the anatomy of the nasal cavity and the chemical characteristics of the drug [1].

Drug bioavailability can be affected by physiological barriers such as mucociliary clearance, the pathophysiology of the nasal mucosa, nasal metabolism, presence of mucus, and the low permeability of the blood-brain barrier. Moreover, drug bioavailability can be modified by its physicochemical characteristics such as solubility and dissolution ability, molecular size and weight, lipophilicity, pKa, and the pH of the environment. Furthermore, drug bioavailability depends on features of formulation as drug concentration, doses, osmolality, and dosage form [2,3]. In recent years, different strategies to evade these problems have been studied. A traditional choice is improving drug absorption by adding excipients.

2. Anatomy of Equine Nose

The central nervous system (CNS) is directly connected with the environment by only one anatomical site, the nasal cavity. The nasal cavity is divided into two symmetrical halves by the median septum, a central partition bone and cartilage. The dorsal wall of the nasal cavity is formed by the frontal and nasal bones. The lateral walls include the maxillary bone, the premaxilla, and the perpendicular portion of the palatine bone. The nasal cavity and the cranial cavity are connected through the ethmoid lamina, which seems to be a concave

sieve and where it is the olfactory bulb. Inside each nostril, there are three turbinates (the dorsal, the median, and the ventral turbinate) responsible for conditioning the incoming air (**Figure 1**).

The nasal cavity is irrigated by the major palatine artery, sphenopalatine artery, and branches of ethmoids. It is innervated by the trigeminal nerve, whose branches take chemosensory and thermosensory information from oral, ocular, and nasal mucosa **[4]**.

Behind the vestibular region, the nasal cavity is divided into the respiratory region, which warms and moistures the incoming air, and the olfactory region, where smell receptors are located **[5]**.

The respiratory region is covered with nasal respiratory mucosa, whose surface has ciliated pseudostratified columnar epithelia and is reddish because of its high vascularization. Respiratory region is located in a region near the dorsal turbinate and formed by nonciliated and ciliated columnar cells, basal cells, and goblet cells. Respiratory region is also related to seromucosal and intraepithelial glands. Seromucosal glands produce most of the nasal secretions **[6–8]**.

The olfactory region is covered with specializing olfactory mucosa and is located in the roof of the nasal cavity and, in varying extension, in adjacent lateral and medial walls. The olfactory tissue is often yellow in color because of the pigment inside sustentacular cells **[5,8]**.

The olfactory region in the nasal cavity has a variable extension, depending on the animal species **[5,8]**.

Lamina propria of the olfactory mucosa is in direct contiguity with periosteum subjacent bone. This conjunctive tissue has considerable blood and lymphatics vessels, unmyelinated olfactory nerves, myelinated nerves, and olfactory glands **[8]**.

The olfactory epithelium, as respiratory one, is pseudostratified, but it contains only three types of cells: the olfactory neural cells, sustentacular cells, and the basal cells, lacking goblet cells **[7,8]**.

The olfactory glands (Bowman's glands) are proper to olfactory mucosa; they are branched serous tubuloalveolar glands that send their protein secretion to the olfactory surface through grooves. The constant flow from glands cleans the mucosa from the rest of odoriferous substances detected, so new smells can be perceived continuously as they emerge **[8]**.

The olfactory system is also formed by accessory organs such as the vomeronasal organ. It is located in both nostrils and in the vomer and is connected to the olfactory bulb with a tube. Its surface is covered with olfactory epithelium and neuronal cells. It has an important function in animal behavior **[8]**.

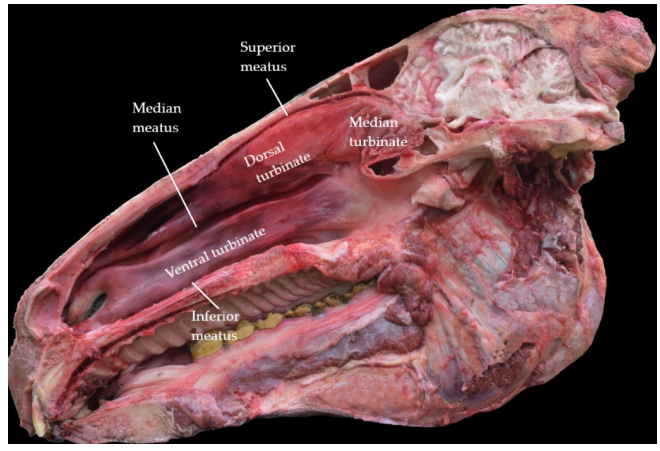

Figure 1: Image of an equine head showing anatomical sites. This figure was originally published in **[4]**.

3. Intranasal Administration

Historically, intranasal administration has been limited to topic application with local effects like antihistaminic and corticosteroids drugs; however, in last years, it has received more attention because it is a promising, suitable, and reliable system for brain drug delivery, focusing on drugs that are inefficient by oral administration or must be administrated with injections.

The intranasal route provides a good surface with easy access avoiding first-pass metabolism. Small and lipophilic drug molecules have better bioavailability because the nasal cavity has easy accessibility to blood capillaries. Moreover, absorption and action of the drug are faster and it has a lower risk of overdosing. The nasal mucosa is more permeable to a wide variety of chemical substances than gastrointestinal mucosa because it lacks pancreatic and gastric enzymes and interference of gastrointestinal content. In addition, it is appropriate for patients with long-term therapy because it is not invasive and avoids the blood-brain barrier; therefore, drug bioavailability in the CNS is increased **[9–12]**. Intranasal administration is one of the most successful approaches for the systemic administration of macromolecules. Its application has been studied in a wide range of indications such as migraine, headache, infection prevention, pain management, hormone replacement therapy, and emergency therapy in seizures. Moreover, small peptides, such as oxytocin (1 kDa), buserelin (1,2 kDa), gonadorelin (1,2 kDa) and nafarelin (1,4 kDa) **[11,12]**, are traded using intranasal formulations.

In equine species, the large surface and irrigation of the nasal cavity allow inferring the viability of this administration route for drug administration **[3]**.

4. Mechanisms of Nasal Drug Absorption

It is important to review the pathways and mechanisms of drug passage from the nasal cavity to the systemic circulation and the CNS to advance intranasal formulations.

The properties of the formulation affecting drug absorption depend on the administration route and the type of dosage form selected. Absorption could be affected by the viscosity and osmolarity of the formulation, the nasal blood flow, the enzymatic activity in the nose, and the physical condition of the nasal mucosa. Also, it is advised to maintain the formulation pH between 4.5 and 6.5 because the pKa value of

the drug can influence the amount of ionized and deionized molecules but also prevent the growth of pathogenic bacteria in the nasal passage, avoid nasal irritation, maintain the functionality of excipients, and sustain the normal physiology of ciliary movement [13].

Firstly, when a drug enters the nasal cavity, it suffers mucociliary clearance in the vestibular region. Therefore, drug molecules move towards the posterior region nasal cavity, where there are in close contact with the respiratory and olfactory region. Then, drug delivery to the nervous system takes place by different paths: olfactory nerve, trigeminal nerve, and lymphatic, vascular, and cerebrospinal fluid routes. However, mechanisms and pathways which describe drug delivery from the nasal cavity to CNS are not totally comprised. Drugs would follow one specific route or a combination of the mentioned routes [14–18].

In the olfactory nerve route, the drug interacts with olfactory neural cells and crosses the olfactory epithelium. It arrives at the lamina propria, crosses the cribriform plate, and reaches the olfactory bulb, being finally directed toward the cerebral tissue or cerebrospinal fluid [19–21].

This route is divided into intraneuronal and extraneuronal routes (**Figure 2**).

In the intraneuronal route, olfactory neural cells catch drug molecules through processes such as endocytosis, passive diffusion, and/or receptor-mediated endocytosis, and the drug reaches the olfactory bulb through axonal transport [3,19].

The intraneuronal route is important but not the predominant route of drug delivery through CNS because the drug needs hours or days to reach the olfactory bulb and the brain. This transport has been reported for gold particles, aluminum salts, and molecules with receptors in olfactory neural cells like wheat germ agglutinin [3,19,22–25].

It has been published that some viruses would use receptor-mediated endocytosis to enter the olfactory epithelium. Many strains of influenza viruses, herpes viruses, and polioviruses use different receptors on the surface of the cilium to enter the olfactory neural cells. Recently, it was published that the SARS-COV-2 virus uses the nose as a gate to the human body. Some articles have shown that the nasal epithelium and olfactory epithelium express angiotensin-converting enzyme II (ACE2), and the virus uses it as an input receptor with cellular serine protease TMPRSS2 for priming protein S. Curiously, loss of smell is an early marker of SARS-COV-2 infection. It is suggested that the damage of sustentacular cells is expressed as nonneural cells and the presence of ACE2 leads to patience with olfactory deficits [11].

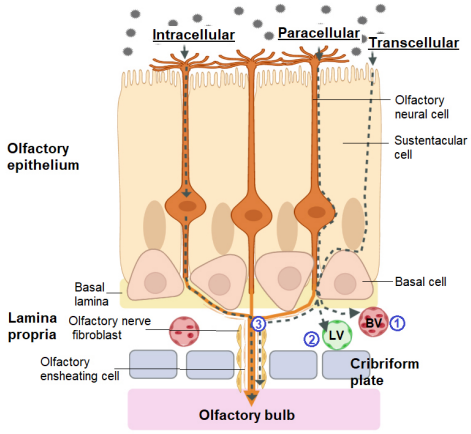

Figure 2: Drugs can take two different routes to cross the olfactory epithelium (intracellular and extracellular route, involving paracellular and transcellular mechanisms). Once the drug reaches the lamina propria, it can (1) be absorbed into the blood vessels and reach the circulation (BV), (2) be absorbed into the lymphatic vessels and drain to the cervical lymph nodes (LV), and (3) travel to the perineural space between the sheathed olfactory cells and the fibroblasts of the olfactory nerve towards the bulb.

In the extraneuronal route, the drug crosses the epithelium by paracellular route between the olfactory neural cells and the basal cells or by transcellular route through basal cells; then, it is transported to the olfactory bulb (**Figure 2**). This mechanism is fast and the drug needs only a few minutes or half an hour to reach the olfactory bulb or other areas in CNS [11,19,21,26].

The majority of publications about intranasal administration report fast drug delivery, high concentrations in CNS, and immediate clinical effects, consistent with the extracellular route [3].

In the trigeminal route, a drug is transported through this nerve; although it has been considered a minor mechanism, in recent years, its importance has been recognized [3].

Olfactory and respiratory epitheliums are innervated by the trigeminal nerve, which reach the CNS at the pons with a small portion ending inside the olfactory bulb. A specific characteristic of the trigeminal nerve is that it enters the brain from the respiratory epithelium through the anterior lacerated foramen near pons and the cribriform plate near the olfactory bulb, therefore making entry points, after intranasal administration, in rostral and caudal areas in the brain. Other nerves that innerve the head and face, such as facial nerve or other sensorial structures in the nasal cavity, can provide entry points to CNS after intranasal administration. First-time trigeminal route in intranasal drug administration was demonstrated, I125-labeled insulin growth factor was used. It showed high radioactivity levels on branches from trigeminal nerves, trigeminal ganglion, the pons, and olfactory bulbs; this is consistent with the transport along the olfactory nerve and trigeminal nerve [19].

Furthermore, the nasal septum contains a little aperture near the base to access the vomeronasal-terminal nerve through the vomeronasal organ. Drugs would be absorbed directly to

the brain through the vomeronasal organ and terminal nerve [18,27,28].

When the drug reaches the olfactory bulb, it is transported to other parts on the brain for diffusion using a perivascular bomb driven by arterial pulsation [21,29].

The nasal mucosa has high blood irrigation and permeability, becoming an interesting administration pathway. Its large surface and vascularity contribute to quickly warming and moisturizing the incoming air; these factors predispose it as a formidable drug absorption site [30,31]. Blood vessel density in respiratory mucosa is higher than that in olfactory mucosa, so respiratory mucosa is ideal for systemic drug absorption.

Blood vessels in the respiratory region contain a mix of continuous and fenestrated endothelium, allowing small and large molecules administered intranasally to enter systemic circulation, reaching CNS passing through the blood-brain barrier. It is expected that small lipophilic drugs get into the bloodstream easier than large or hydrophilic drugs such as peptides or proteins. Also, it is possible that instead of being distributed through the systemic circulation, xenobiotics can enter the venous blood supply in nasal passages where they are rapidly distributed to the carotid artery blood supply, which feeds the brain and spinal cord in a process known as countercurrent transfer [19].

5. Penetration Enhancers

In the development of intranasal formulations, there is a wide range of strategies for increasing drug bioavailability. A mechanism that allows achieving this objective is adding excipients to improve drug penetration and absorption. Pharmacopeia defines excipient as any substance added intentionally to the formulation of any dosage form, which is different from the active pharmaceutical ingredient. The term absorption enhancer refers to an agent whose function is to increase absorption by enhancing membrane permeation rather than increasing solubility, therefore such agents are sometimes more specifically named permeation enhancers [1,32,33].

Penetration enhancers have been studied to improve the efficacy of peptides, proteins, and other active pharmaceutical ingredients with an inefficient permeability. Penetration enhancers are functional excipients added to formulations to improve the permeation of active pharmaceutical ingredients through biological barriers [34].

An ideal penetration enhancer has some features: at the concentration used, it is pharmacologically inert; its effect must be specific, appropriate with predictable duration, should not cause irritation, allergy, or toxicity, and effective at small concentrations. Also, it should be compatible with the drug and other adjuvants present in the formulation and be able to abide in contact with the nasal mucosa time enough to extend its maximum effect; it should not have an unpleasant smell or taste and should not harm the membrane [1,30,35].

The mechanisms of action of penetration enhancers are not totally understood. However, it is supposed they act by exerting one or both of the following effects [30,36]:

- Physicochemical effects: some penetration enhancers can alter drug physicochemical features by modifying drug solubility, drug partition coefficient, or weak interactions with the drug. These mechanisms are observed at low concentrations.

- Membrane effects: many penetration enhancers work acting on the surface of the nasal membrane. Although most of these enhancers are not harmful, it is necessary to determine the extension and consequences. In most cases, penetration enhancers have a transitory effect without irreversible consequences.

In the pharmaceutical industry, there is a wide range of penetration enhancers. Behl *et al.* [30] have recognized the difficulty of precisely classifying these agents in a meaningful manner because they often have overlapping chemical properties, and their penetration mechanisms are not wholly understood. In this review, classification proposed by Behl *et al.* [30] is applied.

5.1. Cyclodextrins

In the last few years, cyclodextrins have become a focal point of investigation in different areas of applications, with special interests of the pharmaceutical industry. Most intranasal formulations contain penetration enhancers for improving absorption of lipophilic drugs, increasing their aqueous solubility and/or improving their nasal penetration [36–38].

Cyclodextrins are cyclic oligosaccharides of six or more α-d- glucopyranoses units joined with 1-4 glycoside bonds, produced by the action of enzymes on starch [39,40].

Although they were described in 1891, it was not until 1976 that they were used as part of a pharmaceutical product of Ono Pharmaceutical Co. company [41,42].

The commonest natural cyclodextrins (α-cyclodextrin, β-cyclodextrin, and γ-cyclodextrin) are formed by 6, 7, and 8 glucose units.

Cyclodextrins derivatives are obtained for the substation of hydroxides group instead of desirable functional groups. Nowadays, more than 1,500 different cyclodextrins derivatives have been described, but only a few are available as pharmaceutical excipients because the analysis of toxicities is costly. It is unlikely that a new derivative would give a wide advantage over the currently employed cyclodextrins; therefore, the availability of new cyclodextrins may increase very slowly. Pharmaceutical products often have methyl, hydroxypropyl, and sulfobutylether derivatives to improve their solubility and capability of inclusion above natural cyclodextrins [42–44].

Cyclodextrins act as a solubilizer and permeation enhancer for nasal drug delivery. Cyclodextrins interact with poorly water-soluble substances increasing their apparent solubility. The mechanism of solubility is given for the formation of an incursion complex where the guest molecule (drug) and host molecule (cyclodextrin) are in a dynamic balance with the complex [1,38].

As a result of chair conformation glucopyranose units, cyclodextrins take a cone truncated shape (**Figure 3(a)**). The hydroxyl residues are oriented outside the cone, with the primary hydroxyl groups of the sugar in the narrowest part of the cone and the secondary hydroxyl group in the widest part. The central part of the cavity from the cyclodextrin molecule

is aligned with the carbon skeleton and glycoside oxygen of glucose. The ends of cyclodextrins that have a hydroxyl group are polar and their interiors are relatively nonpolar [36,38].

In an aqueous solution, the cyclodextrin cavity is slightly nonpolar and is occupied by water molecules that are unfavorably energetic for the formation of a polar–nonpolar interaction, so they can be easily replaced with an appropriate "guest molecule" less polar than water. The dissolved cyclodextrin is the host molecule and part of the driving force of complex formation is the substitution of water molecules with high enthalpy for the adequate guest molecule. Cyclodextrins are attached to guest molecules for the van der Waals bond, hydrogen bond, hydrophobic bond, and covalent bond. One, two, and three cyclodextrins molecules have one or two guest molecules. This mechanism is known as molecular encapsulation (**Figure 3(b)**). The most frequent and simple situation is when the guest: host relationship is 1:1, although there could be, simultaneously, more complex associations and other balances could exist with a high order. The complex balance depends on cyclodextrin molecular concentration, guest molecule concentration, host molecule concentration, and water concentration. The bond can be broken with changes in pH and solution temperature [45,46].

The mechanism of cyclodextrins as drug penetration enhancers occurs in specific conditions and is not totally elucidated nowadays. It has been reported that methylated β-cyclodextrin increases transcellular and paracellular movements of peptidic drugs, improving transmucosal absorption. Also, it has been shown that tetradecyl-β-maltoside-cyclodextrin acts by opening tight junctions between cells [12,34].

Natural cyclodextrins and their derivatives are listed as accepted excipients for FDA (The United States Food and Drug Administration). Cyclodextrins toxicity depends on the administration route. When they are orally administered, they are well tolerated and nontoxic; however, when administered through the intravenous route, they become unsafe since they can crystallize. Cyclodextrins can be corrosive on tissues; however, methylated β-cyclodextrin and dimethylated β-cyclodextrin do not show this local effect and do not produce local nasal toxicity. In fact, the local nasal irritation of methylated β-cyclodextrin is lower than that of benzalkonium chloride [38,43].

Cyclodextrins have been used as intranasal penetration enhancers for calcitonin, insulin, glucagon, and recombinant granulocyte colony-stimulating factor. In the market, there is a commercial cyclodextrin product called KLEPTOSE® HPB (hydroxypropyl-β-cyclodextrin), which is proposed as an attractive excipient for intranasal drug administration due to its capacity as a drug solubilizer and low toxicity [34,37].

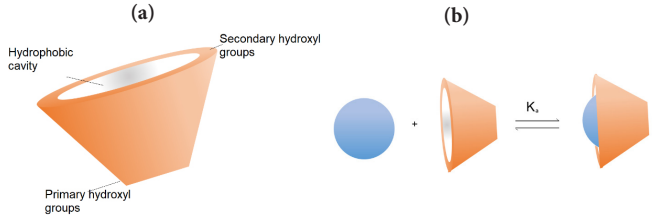

Figure 3(a): Descriptive scheme of molecular structures of cyclodextrins. Cyclodextrins have a truncated shape where their cavity is lipophilic. Hydroxylic groups are outward-oriented, where hydroxyls are in a wider zone and primary hydroxyls in a narrower zone [40]. **(b):** Simplified graphic of molecular encapsulation. The guest lipophilic molecules (circle) interact with cyclodextrin (host molecule) with van der Waals bonds, hydrogen bond, and hydrophobic bond, moving the water, because they are entropically favored for their lipophilic nature.

5.2. Chitosan and Derivatives

Chitosan is cationic linear polysaccharides obtained from the deacetylation of chitin, the second most abundant polysaccharide in nature after cellulose; it is present in crustacean shells (shrimp and crabs), the exoskeleton of insects, and fungal cell wall [47,48].

Chitosan is a copolymer composed of 2-acetamido-2-deoxy-β-D-glucopyranose and 2-amino-2-deoxy-β-D-pyranose units. It has a primary amine group and three free hydroxyls groups for each unit of C_6 (**Figure 4**). This chemical substance is not usually defined as an exact composition, but it is characterized by the number of sugar units of each polymer molecule and deacetylation grade. These proprieties impact biological and physical polymer factors [48–50].

Chitosan and derivatives are known as versatile materials because they have diverse activities; they are biocompatible, biodegradable, nontoxic, cheap, and, usually, nonallergenic. Moreover, they have superior physical properties such as large superficial area, porosity, conductivity, and being easily malleable in different shapes, such as films, sponges, fibers, gel, beads, nanoparticles, microparticles, powders, and solutions. These characteristics have led to chitosan and its derivatives being used in numerous agricultural, biomedical, and pharmaceutical applications. Applications in the biomedical industry include wound healing, genetic engineering, water treatment, implant preparation, tissue engineering, obesity treatment, and dietary supplements to lower cholesterol. Moreover, since they have antibacterial activity, they have been used in antibacterial food and textile packaging materials. They also have antiulcer, antioxidant, antifungal, anti-HIV, and antitumor activity. On the other hand, chitosan is used in the pharmaceutical industry as an effective penetration enhancer for hydrophilic drugs with poor absorption and as a drug carrier, not only improving their absorption but also stabilizing their components for targeting and improving their liberation. Absorption improvement through the intestinal and nasal epithelia has been reported for calcitonin,

insulin, and buserelin. Pharmaceutical formulations based on chitosan are presented in **Table 1** [47,49,52,53].

Regarding chitosan derivatives, applications of N-trimethyl chitosan chlorides, esters, and conjugates have been reported.

N-trimethyl chitosan chlorides (TMC) are quaternary chitosan derivatives with higher water solubility, intestinal permeability, and absorption over a wide pH range. The polymer of TMC is designated as TMC-20%, TMC-40% and TMC-60% according to its degree of methylation. It has been observed that the higher the substation degree, the lower its solubility. Hamman *et al.* have shown that quaternization of TMC decreases the transepithelial electrical resistance and thereby influences drug absorption-enhancing properties [54].

Chitosan-derived esters (glutamate, succinate, phthalate) have different solubility profiles. Chitosan-derived esters-based matrix has been successfully used in many formulations, such as sodium diclofenac with a specific delivery to the colon [52].

Lastly, chitosan can be conjugated to bioactive excipients for delivering active ingredients, such as calcitonin. Some chitosan conjugates, such as 5- methylpyrrolidine chitosan and chitosan-4-thiobutylamidine, have shown enhanced penetration and mucoadhesive properties. Guggi and Bernkop conjugated an enzyme inhibitor with chitosan, resulting in a polymer that prevents drug enzymatic degradation by trypsin and chymotrypsin, concluding that these conjugated chitosans have a promising peptide drug delivery as calcitonin [52].

Table 1: Formulations based on chitosan (type of system and drugs) [52].

Types of system	Drug
Tablet	Diclofenac sodium theophylline, mesalamine, glipizide
Capsules	Insulin
Microspheres/microparticles	Gentamicin sulfate, hemoglobin, diclofenac, clarithromicin, propranolol-HCl famotidine, bovine serum albumin, clozapine
Nanoparticles	Gadopentetic acid, bovine serum albumin, ascorbic acid, cyclosporin A, doxorubicin
Beads	Bovine serum albumin, insulin
Films	Ofloxacin, paclitaxel
Gel	5-Fluorouracil

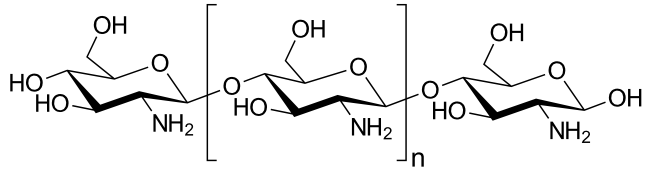

Figure 4: Scheme of the molecular structure of chitosan. Figure adapted from [51].

It has been reported that chitosan is applicable to the intranasal formulation for proteins delivery. Chitosan has mucoadhesive proprieties, probably generated for ionic interaction between positively charged amino groups in chitosan and negatively charged sialic acid residues in mucus. It has demonstrated that the strong cationic nature of chitosan promotes interaction with sialic acid present in the mucosa; hence, it opens the tight junctions of nasal epithelial cells. Intranasal formulation with chitosan could improve drug time residence in tissues and cells, showing a sustained drug release, increasing drug bioavailability, and minimizing administration frequency. Also, it has been found that chitosan improves drug penetration through mucosa without damage to the biological system. Illum *et al.* demonstrated that chitosan glutamate can enhance the passage of insulin through rat and sheep nasal mucosa. Histological examination of rat nasal treated with 0.5% (p/v) of chitosan glutamate during 60 minutes revealed slight structural changes [1, 50, 52].

The efficacy of chitosan as a permeation enhancer was confirmed using salmon calcitonin in rats. The absolute bioavailability of calcitonin was higher in animals that were treated with 1% free amine than rats that received 5% of dimethylated β-cyclodextrins. It was found that free amines and chitosan salts produce mild nasal irritation. Based on these results, chitosan seems to be a safe and effective enhancer for intranasal administration [49,50,52,55].

5.3. Surfactants

Surfactants are amphiphilic molecules with a hydrophilic head and hydrophobic tail, forming a polar asymmetric structure (**Figure 5**). Surfactants can decrease the surface tension between air and water phases, improving the miscibility of substances allowing their diffusion or mixing as an emulsion in water or other solvents [55,56].

These chemicals have applications in the food, petrochemical, and pharmaceutical industries. In the latter, they are used for drug solubility in an aqueous medium, as emulsion components, for vehicles of formulations for the oral or transdermal route, as a plasticizer in release system for drugs with semisolid form, and as drug penetration enhancer or absorption enhancer [57–59].

When surfactants are added in an aqueous medium, they reduce the surface tension of water. As surfactant concentration increases, surface tension falls to a certain concentration where surfactants molecules form micelles spontaneously. The hydrophobic zone of the molecule is sequestered from an aqueous medium, which is highly polar, through a surrounding layer, and depending on the surfactant, it can take a cylindrical, spherical, hexagonal, laminar cubic, inverted cylindrical, or inverted spherical shape. When these micelles are formed, the addition of an excess of surfactant has no effect on the surface tension of water. This concentration in

which surface tension is constant is known as critical micellar concentration (CMC) [57,60].

Surfactants can be natural or synthetic, soluble or insoluble, and their chemical structures can be simple or polymeric. Depending on the molecule charge, they are classified into anionic, cationic, nonionic, and zwitterionic surfactant [56].

Anionic surfactants have a negative charge in the hydrophilic region of the molecule. Their hydrophilic groups are classified into phosphate ester (such as aryl ether phosphates and acyl ether phosphates), fatty acid salt type, salt type sulphonate (for example, sodium dioctyl sulfosuccinate or alkylbenzene sulfonates), and carboxylic acid. They can be used as foaming agents, antistatic agents, dispersants, detergents, and emulsifiers (**Figure 6(a)**) [56,58].

Cationic surfactants have a positive charge in the hydrophilic region of the molecule, for example, an amine with a long chain or a quaternary ammonium salt such as dimethyldioctadecylammonium chloride or benzalkonium chloride. They are classified into open chain cationic surfactants, heterocycle group cationic surfactants, and cationic surfactants with intermediate linkage. They are used in corrosion, oxidation, sterilization, and cleaning skin wounds. They also have bactericidal activity in a wide range of Gram-positive and certain Gram-negative microorganisms; this is the principal reason for their use in the pharmaceutical industry (**Figure 6(b)**) [57,58].

Nonionic surfactants have no charge on the head of the molecule. They are classified into polyoxyethylene esters, poloxamers, and polyol esters. The latter include glycol, glycol esters, and sorbitan derivatives. Sorbitan fatty acid esters (Spans) and their ethoxylated derivatives (called Tweens) are the most widely used nonionic surfactant. Nonionic surfactants are used in the paper, textile, food, and plastic industry and in the production of glass and pesticides and are widely used in the development of pharmaceutical formulations **(Figure 6(c))** [56,58].

Zwitterionic or amphoteric surfactants have a positive and negative charge in their molecule. The most common amphoteric surfactants are betaine derivatives. Depending on the pH of the surrounding medium, surfactants acquire positive, negative, or null charge: in solution with acid pH, they have a positive charge, while if they are in solutions with alkaline pH, they have a negative charge and in solution with a pH in the isoelectric point, both groups have equal ionization. Amphoteric surfactants have minimal toxicity and they have an excellent biodegradation. They are in shower gels, shampoo, cosmetics, industrial softeners, and antistatic agents **(Figure 6(d))** [57,58,61].

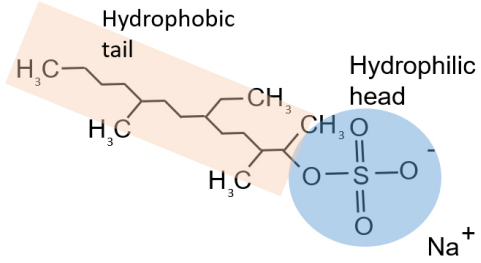

Figure 5: General molecular structure of surfactants. Figure adapted from [57].

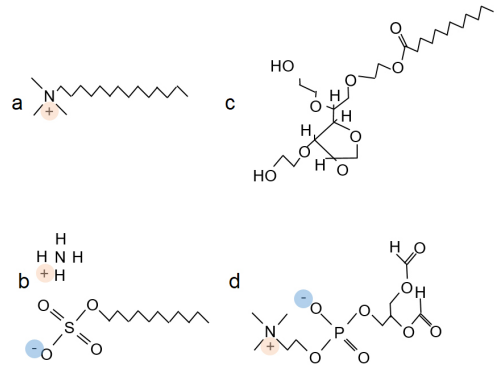

Figure 6(a): Scheme of the molecular structure of cationic surfactant (cetrimide). They have a positive charge in the hydrophilic region of the molecule. Figure adapted from [62]. **(b):** Scheme of the molecular structure of anionic surfactant (ammonium dodecyl sulfate). They have a negative charge in the hydrophilic region of the molecule. Figure adapted from [63]. **(c):** Scheme of the molecular structure of nonionic surfactant (Tween 20), which carries no charge. Figure adapted from [64]. **(d):** Scheme of the molecular structure of zwitterionic surfactant (egg phospholipids). Figure adapted from [65].

The mechanism of action has a thermodynamic origin. A surfactant (whose name has the origin in surface-active agent) is a substance present in low concentration inside a system and it can be adsorbed onto the surface or interface and alter the free energy of these surfaces or interfaces. The term interface indicates the union between two or more immiscible phases; the term surface denotes an interface where one could be water and the other, air. Free energy is the minimal amount of necessary work to make this interface. Interfacial free energy per area unit is the measure used to determine the surface tension between two areas. Surface tension is a measure of the difference of nature from two phases which are in an interface (or superficies). The higher the difference of the nature between the two phases, the higher the surface (or interfacial) tension [66].

The minimal work required to make an additional amount of interface is calculated as the product between interfacial tension and the increment of the area in this interface. Surfactants are substances that, in low concentration, adsorbed part or the entire interface in the system, changing significantly the amount of work necessary to expand these interfaces [66].

Table 2 shows some surfactants listed in the FDA`s inactive ingredient guide [67].

Surfactants are the most used excipients in intranasal formulations for improving drug permeability through mucosa

[66]. The addition of surfactant to intranasal formulation could modify the permeability of the nasal membrane, facilitating the absorption of drugs. It has been reported that several surfactants improve drug nasal absorption, increasing drug bioavailability significantly. Some surfactants can alter and even dissolve biological membranes [68].

Moreover, surfactants have been widely used in the aerosol nasal formulation. These substances not only reduce surface water tension but also facilitate the atomization of the formulation. In intranasal spray formulations, the formation of a uniform nebulization depends on the device and the formulation, so the use of surfactants is common to achieve an effective dose (by droplet size and nebulization) [58].

As a result, surfactants have been extensively used in the formulation for their potential to improve intranasal drug absorption (**Table 3**). Li *et al.* studied the potential of some nonionic surfactants as penetration enhancers in the intranasal administration of rats using an *in vitro* perfusion technique. The drug was sumatriptan and nonionic surfactants were sucrose laurate ester, cremophor, and poloxamer 188, being sucrose laurate ester the most effective [68,69].

Velloso *et al.* have recently published an article comparing two surfactants (a cationic surfactant (cetrimide) and a nonionic surfactant (Tween 80)) as penetration enhancers for butorphanol in an intranasal formulation for use in equines. The results showed that, at the concentrations used, Tween 80 was the most suitable for intranasal formulations of butorphanol for equine application [70].

Regarding toxicity, daily exposure to unprotected surfactants is related to a large number of dermatological problems, such as irritation and allergy, in different degrees of severity. Alkyl polyglucosides, zwitterionic surfactant (such as betaines, amido betaines, and isethionates), and various polymeric surfactants are known to be smooth in the skin, while ethoxylated fatty alcohols cause irritation. Long-chain surfactants (greater than 12 carbons) cause maximum skin irritation. Cationic surfactants are more toxic than anionic surfactants, but anionic surfactants are more toxic than nonionic surfactants [61,71].

Surfactants, when used as an enhancer in intranasal formulations, can cause nasal irritation, epithelial toxicity, and ciliated activity [1].

On the other hand, chemically synthesized surfactants are mostly derived from petroleum and are not usually biodegradable, leaving a toxic residue in the environment. In recent years, the research and application of biosurfactants have grown. Biosurfactants are defined as surfactants produced by microorganisms from a wide variety of substances. They have unique proprieties such as low toxicity, functionality in extreme conditions, and biodegradability. The large diversity of these molecules supports their potential use in the oil industry, medicine, agriculture, food industry, and cosmetics, among others [59,71].

5.4. Bile Acids, Their Salts, and Derivatives

Bile acids are employed in drug formulation for enhancement drug penetration through mucosa, including oral, nasal, ocular, buccal, pulmonary, and rectal, and through the blood-brain barrier. The mechanism of action as penetration enhancer is based on their capacity to open tight junctions, act over mucolytic activity, and increase the solubility of hydrophobic drugs, increasing the apical and basolateral fluidity and promoting the chemical and enzymatic activity of drugs [12,72–74].

Bile acids are amphiphilic molecules synthesized in the liver from cholesterol; they are the principal substance of bile. They act in the intestine as a biological surfactant on the solubilization and absorption of lipids from dietary fats and endogenous soluble molecules such as cholesterol, fatty acids, and hydrophobic drugs. Beyond their role in digestion, they are also involved in other important physiological processes like glucose homeostasis regulation, lipids metabolism regulation, modulation of inflammatory and immunomodulatory processes, and antibacterial action in the small intestine [72,75,76].

Bile acids have a specific molecular structure due to the presence of a steroid nucleus which is large, rigid, and plane. Its structure consists of four rings, three of them with six carbons and one with five carbons (**Figure 7**). The concave side (α) of steroid skeleton is hydrophilic due to the presence of hydroxyls groups, while the convex side (β) has angular methyl groups making it hydrophobic. This structure makes bile acids very different from ordinary aliphatic surfactants, which are composed of a polar head and nonpolar tail; as a consequence, they have a high surface activity and in water, they can associate themselves forming polymolecular aggregates, such as micelles or liposomes, as long as, their concentration is above CMC. The facial amphiphilicity of bile acids influences the way they are organized in solution and is presumed to play an important role in their ability to promote membrane penetration of polar drugs. Bile salts are classified into three groups depending on their conjugation with amino acids and the degree of hydroxylation, including trihydroxy conjugates (such as taurocholate and glycolate), dihydroxy conjugate (such as glycodeoxycholate, glycochenodeoxycholate, and taurochenodeoxycholic, and unconjugated forms (such as cholate, deoxycholate, and chenodeoxycholate) [40,72,73].

The micelle formation of bile salts allows facilitating the transcellular drug passage, improving drug absorption. In aqueous solutions, the anions of bile salts are joined together, making simple micelles; moreover, they can form mixed micelles when combined with polar lipids, conventional surfactants, or amphiphilic drugs. Mixed micelles usually have lower CMC and better solubility capacity than the individual components due to synergic interactions. Furthermore, mixed micellar systems can be prepared without the help of organic solvents and have been applied in pharmaceutical formulations to improve the bioavailability of poorly water-soluble drugs, such as oxaprozin, rebamipide, and doxorubicin [73–76,78–80].

Bilosomes are different vesicular systems that contain bile salts and liposomes. Incorporating bile salts into liposomes can stabilize the membrane against the noxious effects of physiological bile acids in the gastrointestinal tract and facilitate the internalization of particles. Since bilosomes are produced from natural lipids are biocompatible, they have been used as oral administration systems for poorly water-

soluble drugs, such as peptide drugs or vaccines, due to greater stability to acid pH, enzymes, and bile salts [73,75].

Polymeric nanoparticles constitute a new generation of the therapeutic delivery platform with several advantages over other lipid-based carriers, such as liposomes or micelles. They show a better circulation time and greater accumulation at the site of action because of their small size and better capacity of cellular penetration, which improve the permeability and retention effect. With this, polymer-lipid hybrids obtain from the conjugation of bile acid and hydrophilic and biodegradables polymer constitutes a promising drug delivery. These amphiphilic hybrids can self-assemble in an aqueous medium to form nano-sized micelles with a unique core/shell structure, which are formed by a nucleus of bile salt and stabilized by the hydrophilic crown. The drug load of hybrids depends on nucleus size and the crown, while the drug entrapment is associated with a higher size in the nucleus. Moreover, the CMC increases as increase the length of the polymer that constitutes the crown. Usually, polyethyleneglycol is the preferential polymer for nanocarrier systems because it improves biocompatibility and hydrophobicity and avoids opsonization [72].

Due to their particular structural properties, bile acids are suitable substances to make supramolecular aggregates, such as the previously mentioned and hyperbranched and hydrogels structures. Furthermore, their good availability, low cost, and simple procedures for derivatization make them an attractive block to design new pharmaceutical formulations for the administration of drugs, vaccines, and biomolecules [72].

The use of hydrophobic bile acids as pharmaceutical excipients has some issues because their accumulation and retention have been linked as one of the main causes of liver damage in cholestasis. Furthermore, they have limited clinical use due to their irreversible mucosal damage and ciliotoxicity. Hydrophobicity is decisive in the cytotoxicity of bile acids. Also, it has been reported that bile acids with hydrophobic nature such as deoxycholic acid exert a tumor-promoting effect. Bile acids have the potential to induce cell death acting as nonspecific detergents (membranolytics) and through receptor-mediated interactions. They can promote the generation of reactive oxygen species, which can modify lipids, proteins, and nucleic acids and, eventually, cause apoptosis or cellular necrosis. It was also reported that dihydroxy bile salts are more toxic than trihydroxic bile salts; however, these toxic effects occur specifically when they are present in supraphysiological concentration. On the other hand, it has been reported that hydrophilic bile acids, such as ursodeoxycholic acid, exert a cytoprotective effect and decrease toxic activity of hydrophobic bile acids. It was demonstrated that phosphatidylcholine prevents bile salt toxicity in epithelia and gastrointestinal membrane, this effect can be linked with the formation of less toxic micelles. The relative cytotoxicity of bile acids depends on their structure and membrane properties such as composition, fluidity, charge, and hydrophobicity. Little structural modification of natural bile salts has led to bile acids derivatives with lower toxicity. Due to the high toxicity of some hydrophobic bile acids, many semisynthetic analogs have been developed with better toxicological and pharmaceutical properties. Colylsacorsin is a nontoxic bile acid derivative that has been investigated as an enhancer excipient. This semisynthetic bile acid increased *in vivo* and *in vitro* the permeability and absorption of peptide drugs, octreotide, and desmopressin, but, to a lesser extent compared with chenodeoxycholic acid. However, for other peptidic drugs, such as calcitonin and parathyroids hormones, it has a more favorable toxicological profile. It has been reported that the substitution of hydroxyl groups with keto groups produces bile salts and significantly fewer surfactants that are less lipophilic with less toxicity in the membrane; in this context, sodium desoxycholate and taurocholate cause mild inhibition of mucociliary clearance [72,73,79].

Although some studies have indicated that bile salts can cause nasal irritation when used in a concentration above 0.3%, the concentration should not be a limiting factor for the research in the nasal formulation because there is vast evidence that they have a positive effect on drug absorption by intranasal administration [1,76].

Duchauteu *et al.* studied the effect of six bile salts in the intranasal absorption of gentamicin in rabbits. The enhancers used were cholate, taurocholate, glycocholate, desoxycholate, taurodeoxycholate, and glycodesoxycholate salts. They found that gentamicin without bile salts does not penetrate through the nasal mucosa. Furthermore, they found that the better enhancers were sodium cholate and sodium taurodeoxycholate, and sodium desoxycholate was extremely cytotoxic [81].

Park *et al.* investigated the penetration of acyclovir through nasal mucosa in rats using as enhancer mixed micelles of bile salts-acylcarnitines. The bile salt was sodium glycolate, which increased penetration [82].

Table 2: Surfactants listed in the inactive ingredient guide by FDA [67].

Type of surfactant	Surfactant	Route	Dosage form
Anionic surfactants	Ammonium	Topical	Aerosol, emulsion, suspension, shampoo
		Buccal	Tablet, delayed release
		Dental	Gel, paste, dentifrice
	Sodium lauryl	Oral	Capsule, coated pellets, drops, granule, powder (for suspension), chewable, effervescent, film-coated, tablet, orally disintegrating
		Respiratory	Powder
		Sublingual	Tablet
		Topical	Cream, gel, spray, tablet, shampoo, ointment
Cationic surfactants	Benzalkonium	Auricular (otic)	Solution, suspension
		Intrarticular	Injection
		Intramuscular	Injection
		Nasal	Liquid, solution, spray
		Ophthalmic	Drops, gel, solution, suspension
		Respiratory	Spray, solution
		Topical	Cloth, ointment, drops, solution, shampoo
	Quaternium-	Topical	Cream, lotion
		Auricular (otic)	Drops, solution, suspension
Nonionic surfactants	Polysorbate	Intramuscular	Injection, powder, suspension
		Intravenous	Injection, solution, liquid, solution concentrate
		Nasal	Spray, metered
		Ophthalmic	Drops, emulsion, solution, gel-forming, extended released
		Oral	Capsule, coated, delayed released, liquid-filled, concentrate,granule for suspension, powder, tablet
	Poloxamer	Oral	Suspension
		Topical	Gel, lotion
Zwitterionic surfactants	Egg	Intravenous	Emulsion, injection, powder, lyophilized (for solution)
		Oral	Tablet
	Lecithin, soybean	Oral	Capsule, extended release, suspension, tablet, film- coated
		Respiratory (inhalation)	Aerosol, metered

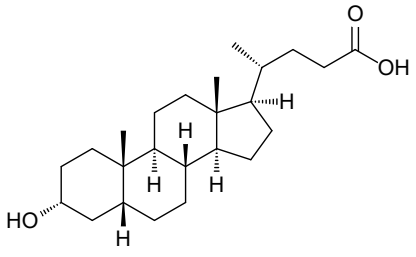

Figure 7: Molecular structure of bile acids. The steroid nucleus is formed by four rings. The concave side (α) is hydrophilic and the convex side (β) is hydrophobic. Figure adapted from [77].

5.5. Sodium Taurodihydrofusidate

Bile salts have limited ability as penetration enhancers through the mucosa, so this fact has stimulated the search for other structurally related substances, which may further mucosal permeability, exhibit greater protein compatibility, and minimize related toxicity and irritation [83].

The sodium salt of fusidic acid was developed in 1962 by LEO Pharmaceuticals (Denmark) from the fermentation of the fungus Fusidium coccineum. The physicochemical proprieties of the sodium fusidic salts and their glycine and taurine conjugates are similar to free bile acids and their conjugates. Sodium taurodihydrofusidate (STDHF) is a semisynthetic derivative that does not exhibit antibacterial activity and it has a similar structure and micellar characteristics to bile salt taurocholate (**Figure 8**) [83–85].

STDHF was widely used as an intranasal penetration enhancer for peptide and protein drugs. It was reported that intranasal bioavailability of peptide and protein drugs such as insulin,

calcitonin, human growth hormone, and octreotide were higher when STDHF was used as a penetration enhancer [1].

Deurloo *et al.* reported that 1% of STDHF increases the bioavailability of insulin in rats and rabbits by intranasal administration [87].

Baldwin *et al.* showed that the bioavailability of human growth hormone by intranasal administration increased 11-fold in rats and rabbits and 21-fold in sheep when 0.5% STDHF was used as an excipient [1,88].

Kissel *et al.* studied absorption improvement and humans' tolerability of octreotide after intranasal administration using STDHF. The bioavailability of formulations with 1.7 and 3% of STDHF were 25.7 % and 29% higher, respectively [89].

It was reported that STDHF, as bile salts, inhibits protease activity and can form transitory pores over lipid membrane [90].

It was reported that STDHF has a mild effect on nasal morphology with a little influence on the ciliary movement. Hermens *et al.* studied the *in vitro* effect of the frequency of ciliary movement of the human nasal mucosa using STDHF as an enhancer [91]. It was evidenced that ciliary movement decreases as the concentration of STDHF increases. When STDHF was used in 0.1 % and 0.2% concentrations, these negative effects decreased, but concentrations of 0.3% or higher induce ciliostasis. However, ciliostasis by STDHF was lower than with laureth-9 (a nonionic detergent) and deoxycholate (a bile salt) [1,90].

Table 3: Some of the surfactants evaluated and drugs whose absorption was enhanced [68].

Surfactant	Drug whose absorption is enhanced
1% sodium taurocholate	Scarlet fever toxin
1% sodium glycocholate	Gentamicin
Polyoxyethylene-9-lauryl ether(BL-9, 0,5%) and sodium glycocholate	Hydralazine
1% sodium deoxycholate	Aerosol spray of insulin
Polysorbate 80 (1%)	Progesterone, Testosterone
Carbopol934	Powder form of insulin
Sodium glycocholate (pH 7.6)	Crystalline porcine insulin administered intranasally

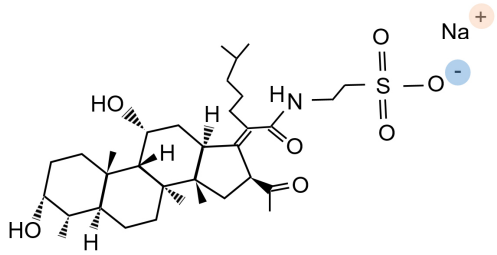

Figure 8: Molecular structure of taurodihydrofusidate. Figure adapted from [86].

STDHF has two very important limitations. First, the tendency to promote the aggregation of proteins and peptides at pHs below their isoelectric points results in dramatic decreases in protein solubility. This is a result of ion pairing between the negatively charged sulfonic acid moiety on STDHF and basic functionalities on the protein. The second limitation is the wide variation in absorption enhancement for different proteins. These limitations decrease the overall utility of STDHF as a nonspecific mucosal permeation enhancer [82].

5.6. Phospholipids

Phospholipids are surfactant compounds found in both animal cells and plant cells and have been explored as nasal permeation enhancers [1].

These substances are lipids that contain phosphorous, a hydrophilic head and a hydrophobic tail. The hydrophilic head and the hydrophobic tail are attached to an alcohol. Depending on the chemical nature of the alcohol, they can be classified into glycerophospholipids and sphingomyelins [92].

Glycerophospholipids have glycerol in their backbone. The sn-1 and sn-2 positions of the glycerol backbone are esterified with fatty acids of varying lengths and degrees of saturation, while the sn-3 position is esterified with phosphoric acid (**Figure 9**). Variation in the head group leads to different glycerophospholipids, such as phosphatidylcholine, phosphatidylethanolamine, phosphatidylglycerol, phosphatidylinositol, or phosphatidylserine. On the other hand, in the backbone of glycerophospholipids, there is glycerol; in the backbone of sphingomyelins, there is sphingosine. Furthermore, phospholipids are classified into natural and synthetic origins [90,93].

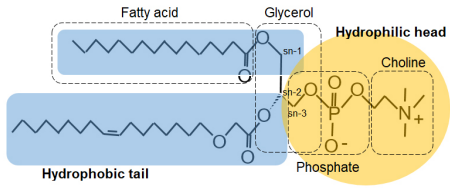

Figure 9: Molecular structure of phospholipids. They are formed by a hydrophilic head and a hydrophobic tail. Glycerophospholipids have glycerol in their backbone and sphingomyelins have sphingosine. The sn-1 and sn-2 positions of the glycerol backbone are esterified with fatty acids with different lengths and degrees of saturation, while the sn-3 position is esterified with phosphoric acid. Variation in the head group leads to different glycerophospholipids, such as phosphatidylcholine, phosphatidylethanolamine, phosphatidylglycerol, phosphatidylinositol, or phosphatidylserine. Figure adapted from [92].

Phospholipids are substances that can self-assemble in an aqueous medium and form colloidal structures, such as liposomes, emulsions, or lipid microspheres. These structures have been widely studied because of their small size and big capacity to take hydrophobic and hydrophilic drugs (**Figure 10**). Also, phospholipids have moisturizing and emulsifying characteristics, which allow them to establish emulsions and cover crystal surfaces to improve the hydrophilicity of hydrophobic drugs. On the other hand, phospholipids are linked to many physiological processes, which provide low toxicity, so they can be used in many administration routes. All these features turn them into potential drug carriers and have been used in many pharmaceutical formulations. In the last few years, various formulations based on phospholipids with clinical applications have been created, such as Doxil®, Cleviprex®, Valium®, and Silybin Phytosome **[92–94]**.

Liposomes have served as carriers for vaccines, antifungals, antitumor drugs, analgesics, and gene therapy drugs. **Table 4** shows some liposome formulations approved for therapeutic use **[93]**.

Regarding the use of phospholipids in intranasal formulations, lysophosphatidylcholine is the most extensively studied penetration enhancer. The absorption of biosynthetic human growth hormone was studied using various permeation enhancers in rats. The relative bioavailability of 25.8% was observed with lysophosphatidylcholine, whereas other permeation enhancers required high concentration to show a similar effect. Drejer *et al.* studied the use of a phospholipid (dodecanoyl-L-α-phosphatidylcholine) as an enhancer for intranasal administration of insulin compared to the subcutaneous route. They found that using this excipient, the intranasal administration was fast, with bioavailability 8.3% higher than subcutaneous administration and without irritation. Alsarra *et al.* studied liposomes of dipalmitoylphosphatidylcholine and cholesterol as an enhancer for intranasal administration of acyclovir by hydrogel in rabbits. This mucoadhesive-liposomal system showed an improvement on acyclovir permeability by intranasal administration **[1,96,97]**.

Furthermore, in the last few years, nanoparticles have been developed based on phospholipids as a strategy to overcome nasal barriers. Natsheh *et al.* designed the first carrier with vesicles based on phospholipid and modified with ethanol and ethyleneglycol for intranasal administration targeting the brain and systemic circulation. Drugs that used this carrier were efficient for treating diseases such as pain, multiple sclerosis, inflammation, migraine, and insomnia in some animal species. After that, Touitou and Natsheh presented phospholipid magnesome, a carrier composed of a phospholipid, propylene glycol, magnesium ion, water, and optionally a mucoadhesive polymer such as alginate salt. It was designed to administer peptides, proteins, and small molecules to the brain; experimental evidence shows that it allows a fast and direct route to the brain **[98]**.

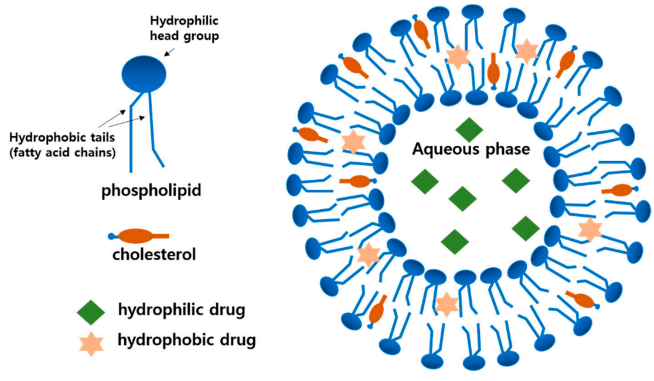

Figure 10: Scheme of liposome structure. They have a small size and they can take hydrophilic and hydrophobic drugs. Figure was originally published in **[95]**.

Table 4: Approved liposomal formulations (commercial name, phospholipids, drug, and year approved [93]).

Product	Phospholipids	Drug	Year approved
Ambisome®	Dihydrogenated Soy Phosphatidylcholine and 1,2-Distearoylphosphatidylglycerol	Amphotericin B	1990 (Europe) 1997 (USA)
Doxil®	Dihydrogenated Soy Phosphatidylcholine and 1,2-distearoyl-sn-glycero-3-phosphorylethanoamine- PEG_{2000}	Doxorubicin	1995 1999 2003 (Europe, Canada) 2007
DaunoXome®	Distearoylphosphatidylcholine	Daunorubicin	1996 (Europe) 1996 (USA)
Myocet™	Egg phosphatidylcholine	Doxorubicin	2000 (Europe)
DepoDur™	1,2-dioleoyl-sn-glycero-3-phosphocholine and 1,2-dipalmitoyl-sn-glycero-3-phosphorylglycerol	Morphine sulfate	2004
DepoCyt®	1,2-dioleoyl-sn-glycero-3-phosphocholine and 1,2-dipalmitoyl-sn-glycero-3-phosphorylglycerol	Cytosine Arabinoside	1999
Lipo-Dox®	Distearoylphosphatidylcholine and 1,2-distearoyl-sn-glycero-3-phosphorylethanoamine-PEG_{2000}	Doxorubicin	2001 (Taiwan)
Marqibo®	Egg yolk sphingomyelin	Vincristine	2012 (USA)

6. Conclusions

The intranasal administration is recognized as a promising route for the systemic and cerebral administration of drugs.

In veterinary medicine, intranasal administration would be an alternative route to intravenous, subcutaneous, oral, or rectal administration. However, this administration route is not commonly used in horses except for local treatments.

Nevertheless, the drug deals with barriers inherent to the anatomy of the nasal cavity and the chemical nature of the drug before it can reach the site of action, which makes the design of intranasal administration formulations a challenge. This review described the most relevant penetration enhancers that would help us overcome this challenge.

On the other hand, nowadays, there is not enough information about the development of intranasal formulations for equines, and considering the extensive advantages of this route, it is expected that, in the future, studies in this area will increase.

Authors' Contributions

Concept – F. L.; Design – M. I. V., F. L.; Supervision – F. L.; Literature Search – M. I. V.; Writing – M. I. V.; Critical Reviews – F. L.

Acknowledgement

All copyright regulations related to reused materials have been followed, whether by referencing the original source or obtaining the proper permissions.

Conflicts of Interest

The authors declare that there is no conflicts of interest.

References

[1] Kim D-D. In Vitro Cellular Models for Nasal Drug Absorption Studies. 1st ed., California, USA: 2008, p. 216–34. https://doi.org/10.1007/978-0-387-74901-3_9.

[2] Bhise SB, Yadav AV, Avachat AM, Malayandi R. Bioavailability of intranasal drug delivery system. Asian Journal of Pharmaceutics (AJP): Free Full Text Articles from Asian J Pharm 2014;2. https://doi.org/10.22377/ajp.v2i4.203.

[3] Muñoz-Cernada A, Fernández-Cervera M, García-Rodríguez JC. Factors involved in the design of nasal delivery systems for peptides and proteins. Biotecnología Aplicada 2013;30:88–96.

[4] Ferreira V, Velloso MI, Vita M, Landoni MF. Vía intranasal: una alternativa para la administración de fármacos de acción central en equinos. Intranasal drug delivery: an alternative for the administration of central acting drugs in horses 2019;39, no. 1. https://doi.org/10.24215/15142590e033.

[5] Singh AK, Singh A, Madhv NVS. Nasal Cavity, a Promising Transmucosal Platform for Drug Delivery and Research

Approaches from Nasal to Brain Targetting. Journal of Drug Delivery and Therapeutics 2012;2. https://doi.org/10.22270/jddt.v2i3.163.

[6] Tortora GJ, Derrickson B, Tzal K, Gutierrez M de los A, Klajn D. Principios de Anatomía y Fisiología. Mexico: México Editorial Médica Panamericana; 2013.

[7] Lochhead JJ, Thorne RG. Intranasal Drug Delivery to the Brain. In: Hammarlund-Udenaes M, de Lange ECM, Thorne RG, editors. Drug Delivery to the Brain: Physiological Concepts, Methodologies and Approaches, New York, NY: Springer; 2014, p. 401–31. https://doi.org/10.1007/978-1-4614-9105-7_14.

[8] Ross MH, Wojciech P, Alday A, Giacomucci M. Histología: texto y atlas : correlación con biología celular y molecular. 7th ed. Barcelona, España: Wolters Kluwer; 2016.

[9] Alnasser S. A Review on Nasal Drug Delivery System and Its Contribution in Therapeutic Management. Asian Journal of Pharmaceutical and Clinical Research 2019:40–5. https://doi.org/10.22159/ajpcr.2019.v12i1.29443.

[10] Dufes C, Olivier J-C, Gaillard F, Gaillard A, Couet W, Muller J-M. Brain delivery of vasoactive intestinal peptide (VIP) following nasal administration to rats. International Journal of Pharmaceutics 2003;255:87–97. https://doi.org/10.1016/S0378-5173(03)00039-5.

[11] Keller L-A, Merkel O, Popp A. Intranasal drug delivery: opportunities and toxicologic challenges during drug development. Drug Deliv and Transl Res 2021. https://doi.org/10.1007/s13346-020-00891-5.

[12] Mujawar N, Ghatage S, Navale S, Sankpal B, Patil S, Patil S. Nasal Drug Delivery: Problem Solution and Its Application. Journal of Current Pharma Research 2014;4:1231–45. https://doi.org/10.33786/JCPR.2014. V04I03.008.

[13] Ozsoy Y, Güngör S. Nasal route: an alternative approach for antiemetic drug delivery. Expert Opin Drug Deliv 2011;8:1439–53. https://doi.org/10.1517/17425247.2011.607437.

[14] Geneser F, Bruel A, Christensen EI, Tranum-Jensen J, Qvortrup K. Geneser Histología. 4th ed. Buenos Aires, Argentina: Editorial médica panamericana; 2015.

[15] Maher S, Casettari L, Illum L. Transmucosal Absorption Enhancers in the Drug Delivery Field. Pharmaceutics 2019;11:339. https://doi.org/10.3390/pharmaceutics11070339.

[16] Williams OW, Sharafkhaneh A, Kim V, Dickey BF, Evans CM. Airway Mucus. Am J Respir Cell Mol Biol 2006;34:527–36. https://doi.org/10.1165/rcmb.2005-0436SF.

[17] Gänger S, Schindowski K. Tailoring Formulations for Intranasal Nose-to-Brain Delivery: A Review on Architecture, Physico-Chemical Characteristics and Mucociliary Clearance of the Nasal Olfactory Mucosa. Pharmaceutics 2018;10:116. https://doi.org/10.3390/pharmaceutics10030116.

[18] Erdő F, Bors LA, Farkas D, Bajza Á, Gizurarson S. Evaluation of intranasal delivery route of drug administration for brain targeting. Brain Research Bulletin 2018;143:155–70. https://doi.org/10.1016/j.brainresbull.2018.10.009.

[19] Dhuria SV, Hanson LR, Frey WH. Intranasal delivery to the central nervous system: Mechanisms and experimental considerations. JPharmSci 2010;99:1654–73. https://doi.org/10.1002/jps.21924.

[20] Agrawal M, Saraf S, Saraf S, Antimisiaris SG, Chougule MB, Shoyele SA, *et al*. Nose-to-brain drug delivery: An update on clinical challenges and progress towards approval of anti-Alzheimer drugs. Journal of Controlled Release 2018;281:139–77. https://doi.org/10.1016/j.jconrel.2018.05.011.

[21] Mittal D, Ali A, Md S, Baboota S, Sahni JK, Ali J. Insights into direct nose to brain delivery: current status and future perspective. Drug Delivery 2014;21:75–86. https://doi.org/10.3109/10717544.2013.838713.

[22] Balin BJ, Broadwell RD, Salcman M, El-Kalliny M. Avenues for entry of peripherally administered protein to the central nervous system in mouse, rat, and squirrel monkey. Journal of Comparative Neurology 1986;251:260–80. https://doi.org/10.1002/cne.902510209.

[23] Baker H, Spencer RF. Transneuronal transport of peroxidase-conjugated wheat germ agglutinin (WGA-HRP) from the olfactory epithelium to the brain of the adult rat. Exp Brain Res 1986;63:461–73. https://doi.org/10.1007/BF00237470.

[24] Kristensson K, Olsson Y. Uptake of exogenous proteins in mouse olfactory cells. Acta Neuropathol 1971;19:145–54. https://doi.org/10.1007/BF00688493.

[25] Perl D, Good P. Uptake of Aluminium into Central Nervous System along Nasal-Olfactory Pathways. The Lancet 1987;329:1028. https://doi.org/10.1016/S0140-6736(87)92288-4.

[26] Westin UE, Boström E, Gråsjö J, Hammarlund-Udenaes M, Björk E. Direct Nose-to-Brain Transfer of Morphine After Nasal Administration to Rats. Pharm Res 2006;23:565–72. https://doi.org/10.1007/s11095-006-9534-z.

[27] Gizurarson S. Anatomical and Histological Factors Affecting Intranasal Drug and Vaccine Delivery. Current Drug Delivery n.d.;9:566–82.

[28] Monti-Bloch L, Jennings-White C, Dolberg DS, Berliner DL. The human vomeronasal system. Psychoneuroendocrinology 1994;19:673–86. https://doi.org/10.1016/0306-4530(94)90049-3.

[29] Ying W. The nose may help the brain: intranasal drug delivery for treating neurological diseases. Future Neurology 2008;3:1–4. https://doi.org/10.2217/14796708.3.1.1.

[30] Behl CR, Pimplaskar HK, Sileno AP, Xia WJ, Gries WJ, deMeireles JC, *et al*. Optimization of systemic nasal drug delivery with pharmaceutical excipients. Advanced Drug Delivery Reviews 1998;29:117–33. https://doi.org/10.1016/S0169-409X(97)00064-1.

[31] Aulton ME. Farmacia: La Ciencia y Diseño de Las Formas Farmacéuticas. 2nd ed. Elsevier España; 2004.

[32] Aungst BJ. Absorption Enhancers: Applications and Advances. AAPS J 2012;14:10–8. https://doi.org/10.1208/s12248-011-9307-4.

[33] Villafuerte Robles L. Los excipientes y su funcionalidad en productos farmacéuticos sólidos. Revista mexicana de ciencias farmacéuticas 2011;42:18–36.

[34] Ghadiri M, Young PM, Traini D. Strategies to Enhance Drug Absorption via Nasal and Pulmonary Routes. Pharmaceutics 2019;11:113. https://doi.org/10.3390/pharmaceutics11030113.

[35] Na L, Mao S, Wang J, Sun W. Comparison of different absorption enhancers on the intranasal absorption of isosorbide dinitrate in rats. International Journal of Pharmaceutics 2010;397:59–66. https://doi.org/10.1016/j.ijpharm.2010.06.048.

[36] Leyva E, Moctezuma E, Leyva R, Oros S. Estudio de los complejos de inclusión de ácido nalidíxico y ácido oxolínico

con ciclodextrinas. Revista de la Sociedad Química de México 2004;48:189–95.

[37] Merkus FWHM, Verhoef JC, Marttin E, Romeijn SG, van der Kuy PHM, Hermens WAJJ, *et al*. Cyclodextrins in nasal drug delivery. Advanced Drug Delivery Reviews 1999;36:41–57. https://doi.org/10.1016/S0169-409X(98)00054-4.

[38] Davis SS, Illum L. Absorption Enhancers for Nasal Drug Delivery. Clin Pharmacokinet 2003;42:1107–28. https://doi.org/10.2165/00003088-200342130-00003.

[39] Connors KA. The Stability of Cyclodextrin Complexes in Solution. Chem Rev 1997;97:1325–58. https://doi.org/10.1021/cr960371r.

[40] Brewster ME, Loftsson T. Cyclodextrins as pharmaceutical solubilizers. Advanced Drug Delivery Reviews 2007;59:645–66. https://doi.org/10.1016/j.addr.2007.05.012.

[41] Villiers A. [On the fermentation of starch by the action of butyric ferment]. Comptes rendus de l'Académie des sciences Sci 1891;112:536–8.

[42] Loftsson T, Duchêne D. Cyclodextrins and their pharmaceutical applications. Int J Pharm 2007;329:1–11. https://doi.org/10.1016/j.ijpharm.2006.10.044.

[43] Muankaew C, Loftsson T. Cyclodextrin-Based Formulations: A Non-Invasive Platform for Targeted Drug Delivery. Basic & Clinical Pharmacology & Toxicology 2018;122:46–55. https://doi.org/10.1111/bcpt.12917.

[44] Popr M. Synthesis of cyclodextrin derivates for practical applications. PhD Thesis. Charles University in Prague, 2016.

[45] Balte AS, Goyal PK, Gejjia SP. Theoretical studies on the encapsulation of Paracetamol in the a, b and g Cyclodextrins. Journal of Chemical and Pharmaceutical Research 2012;4:2391–9.

[46] Szejtli J. Past, present, and future of cyclodextrin research. Pure and Applied Chemistry 2004;76:1825–46.

[47] Cheung RCF, Ng TB, Wong JH, Chan WY. Chitosan: An Update on Potential Biomedical and Pharmaceutical Applications. Marine Drugs 2015;13:5156–86. https://doi.org/10.3390/md13085156.

[48] Schipper NGM, Vårum KM, Stenberg P, Ocklind G, Lennernäs H, Artursson P. Chitosans as absorption enhancers of poorly absorbable drugs: 3: Influence of mucus on absorption enhancement. European Journal of Pharmaceutical Sciences 1999;8:335–43. https://doi.org/10.1016/S0928-0987(99)00032-9.

[49] Dodane V, Vilivalam VD. Pharmaceutical applications of chitosan. Pharmaceutical Science & Technology Today 1998;1:246–53. https://doi.org/10.1016/S1461-5347(98)00059-5.

[50] Illum L, Jabbal-Gill I, Hinchcliffe M, Fisher AN, Davis SS. Chitosan as a novel nasal delivery system for vaccines. Advanced Drug Delivery Reviews 2001;51:81–96. https://doi.org/10.1016/S0169-409X(01)00171-5.

[51] Wikimedia Commons contributors. File:Chitosan chemical structural formula.svg. Wikimedia Commons, the Free Media Repository 2014. https://commons.wikimedia.org/wiki/File:Chitosan_chemical_structural_formula.svg (accessed March 14, 2022).

[52] Bansal V, Sharma P, Sharma N, Pal O, Malviya R. Applications of Chitosan and Chitosan Derivatives in Drug Delivery. Advances in Biological Research 2011;5:28–37.

[53] Shariatinia Z. Pharmaceutical applications of chitosan. Advances in Colloid and Interface Science 2019;263:131–94. https://doi.org/10.1016/j.cis.2018.11.008.

[54] Hamman JH, Schultz CM, Kotzé AF. N-Trimethyl Chitosan Chloride: Optimum Degree of Quaternization for Drug Absorption Enhancement Across Epithelial Cells. Drug Development and Industrial Pharmacy 2003;29:161–72. https://doi.org/10.1081/DDC-120016724.

[55] Aspden TJ, Mason JDT, Jones NS, Lowe J, Skaugrud Ø, Illum L. Chitosan as a Nasal Delivery System: The Effect of Chitosan Solutions on in Vitro and in Vivo Mucociliary Transport Rates in Human Turbinates and Volunteers. Journal of Pharmaceutical Sciences 1997;86:509–13. https://doi.org/10.1021/js960182o.

[56] Suhail M, Janakiraman AK, Khan A, Naeem A, Badshah SF. Surfactants and their role in Pharmaceutical Product Development: An Overview. Journal of Pharmacy and Pharmaceutics 2019;6:72–82.

[57] Sekhon BS. Surfactants: Pharmaceutical and Medicinal Aspects. Journal of Pharmaceutical Technology, Research and Management 2013;1:43–68. https://doi.org/10.15415/jptrm.2013.11004.

[58] Kulkarni VS, Shaw C. Essential Chemistry for Formulators of Semisolid and Liquid Dosages. 1st ed. London, United Kingdom: Academic Press; 1999.

[59] De S, Malik S, Ghosh A, Saha R, Saha B. A review on natural surfactants. RSC Adv 2015;5:65757–67. https://doi.org/10.1039/C5RA11101C.

[60] Schreier S, Malheiros SVP, de Paula E. Surface active drugs: self-association and interaction with membranes and surfactants. Physicochemical and biological aspects. Biochimica et Biophysica Acta (BBA) - Biomembranes 2000;1508:210–34. https://doi.org/10.1016/S0304-4157(00)00012-5.

[61] Tadros T. Chemical Technology of Cosmetics. 1st ed. New Jersey, USA: John Wiley & Sons, Inc; 2013.

[62] National Center for Biotechnology Information. PubChem Compound Summary for CID 14250, Cetrimide 2022. https://pubchem.ncbi.nlm.nih.gov/compound/Cetrimide (accessed March 14, 2022).

[63] National Center for Biotechnology Information. PubChem Compound Summary for CID 16700, Ammonium dodecyl sulfate 2022. https://pubchem.ncbi.nlm.nih.gov/compound/Ammonium-dodecyl-sulfate. (accessed March 14, 2022).

[64] National Center for Biotechnology Information. PubChem Compound Summary for CID 443314, Polysorbate 20 2022. https://pubchem.ncbi.nlm.nih.gov/compound/Tween-20 (accessed March 14, 2022).

[65] National Center for Biotechnology Information. PubChem Compound Summary for CID 10425705, Phospholipids egg n.d. https://pubchem.ncbi.nlm.nih.gov/compound/Phospholipids-egg (accessed March 15, 2022).

[66] Rosen MJ. Characteristic Features of Surfactants. Surfactants and Interfacial Phenomena. 3rd ed., New Jersey, USA:

John Wiley & Sons, Ltd; 2004, p. 1–15. https://doi.org/10.1002/9781118228920.ch1.

[67] Inactive Ingredient Search for Approved Drug Products n.d. https://www.accessdata.fda.gov/scripts/cder/iig/index.cfm (accessed August 25, 2022).

[68] Chhajed S, Sangale S, Barhate SD. Advantageous Nasal Drug Delivery System: A Review. International Journal of Pharmaceutical Sciences and Research 2011;2:1322–30.

[69] Li Y, Li J, Zhang X, Ding J, Mao S. Non-ionic surfactants as novel intranasal absorption enhancers: in vitro and in vivo characterization. Drug Delivery 2016;23:2272–9. https://doi.org/10.3109/10717544.2014.971196.

[70] Velloso MI, Andreeta HA, Landoni MF. Effect of two surfactants on in vitro permeation of butorphanol through horse nasal mucosa. Uniciencia 2021;35:1–9. https://doi.org/10.15359/ru.35-2.1.

[71] Vijayakuma S, Varatharajan S. Biosurfactants-Types, Sources and Applications. Research Journal of Microbiology 2015;10:181–92. https://doi.org/10.3923/jm.2015.181.192.

[72] Pavlović N, Goločorbin-Kon S, Đanić M, Stanimirov B, Al-Salami H, Stankov K, *et al.* Bile Acids and Their Derivatives as Potential Modifiers of Drug Release and Pharmacokinetic Profiles. Frontiers in Pharmacology 2018;9.

[73] Moghimipour E, Ameri A, Handali S. Absorption-Enhancing Effects of Bile Salts. Molecules 2015;20:14451–73. https://doi.org/10.3390/molecules200814451.

[74] Moiseev RV, Morrison PWJ, Steele F, Khutoryanskiy VV. Penetration Enhancers in Ocular Drug Delivery. Pharmaceutics 2019;11:321. https://doi.org/10.3390/pharmaceutics11070321.

[75] Faustino C, Serafim C, Rijo P, Reis CP. Bile acids and bile acid derivatives: use in drug delivery systems and as therapeutic agents. Expert Opinion on Drug Delivery 2016;13:1133–48. https://doi.org/10.1080/17425247.2016.1178233.

[76] Stojančević M, Pavlović N, Goločorbin-Kon S, Mikov M. Application of bile acids in drug formulation and delivery. Frontiers in Life Science 2013;7:112–22. https://doi.org/10.1080/21553769.2013.879925.

[77] Wikimedia Commons contributors. File:Lithocholic acid acsv.svg. Wikimedia Commons, the Free Media Repository n.d. https://commons.wikimedia.org/w/index.php?title=File:Lithocholic_acid_acsv.svg&oldid=637160850 (accessed March 15, 2022).

[78] Bhattacharjee J, Verma G, Aswal VK, Date AA, Nagarsenker MS, Hassan PA. Tween 80–Sodium Deoxycholate Mixed Micelles: Structural Characterization and Application in Doxorubicin Delivery. J Phys Chem B 2010;114:16414–21. https://doi.org/10.1021/jp108225r.

[79] Maestrelli F, Cirri M, Mennini N, Zerrouk N, Mura P. Improvement of oxaprozin solubility and permeability by the combined use of cyclodextrin, chitosan, and bile components. European Journal of Pharmaceutics and Biopharmaceutics 2011;78:385–93. https://doi.org/10.1016/j.ejpb.2011.03.012.

[80] Mukaizawa F, Taniguchi K, Miyake M, Ogawara K, Odomi M, Higaki K, *et al.* Novel oral absorption system containing polyamines and bile salts enhances drug transport via both transcellular and paracellular pathways across Caco-2 cell monolayers. International Journal of Pharmaceutics 2009;367:103–8. https://doi.org/10.1016/j.ijpharm.2008.09.027.

[81] Duchateau GSMJE, Zuidema J, Merkus FWHM. Bile salts and intranasal drug absorption. International Journal of Pharmaceutics 1986;31:193–9. https://doi.org/10.1016/0378-5173(86)90153-5.

[82] Park G-B, Shao Z, Mitra AK. Acyclovir Permeation Enhancement Across Intestinal and Nasal Mucosae by Bile Salt-Acylcarnitine Mixed Micelles. Pharm Res 1992;9:1262–7. https://doi.org/10.1023/A:1015845031488.

[83] Lee WA, Lu HF-L, Maffuid PW, Botet MT, Baldwin PA, Benkert TA, *et al.* The synthesis, characterization and biological testing of a novel class of mucosal permeation enhancers. Journal of Controlled Release 1992;22:223–37. https://doi.org/10.1016/0168-3659(92)90097-B.

[84] Beaudoin M, Carey MC, Small DM. Effects of taurodihydrofusidate, a bile salt analogue, on bile formation and biliary lipid secretion in the rhesus monkey. J Clin Invest 1975;56:1431–41. https://doi.org/10.1172/JCI108224.

[85] Longenecker JP, Moses AC, Flier JS, Silver RD, Carey MC, Dubovi EJ. Effects of Sodium Taurodihydrofusidate on Nasal Absorption of Insulin in Sheep. Journal of Pharmaceutical Sciences 1987;76:351–5. https://doi.org/10.1002/jps.2600760502.

[86] National Center for Biotechnology Information. PubChem Compound Summary for CID 44144474, Sodium taurodihydrofusidate. n.d. https://pubchem.ncbi.nlm.nih.gov/compound/Sodium-taurodihydrofusidate. (accessed March 15, 2022).

[87] Deurloo MJM, Hermens WAJJ, Romeyn SG, Verhoef JC, Merkus FWHM. Absorption Enhancement of Intranasally Administered Insulin by Sodium Taurodihydrofusidate (STDHF) in Rabbits and Rats. Pharm Res 1989;6:853–6. https://doi.org/10.1023/A:1015904404442.

[88] Baldwin PA, Klingbeil CK, Grimm CJ, Longenecker JP. The Effect of Sodium Tauro-24,25-Dihydrofusidate on the Nasal Absorption of Human Growth Hormone in Three Animal Models. Pharm Res 1990;7:547–52. https://doi.org/10.1023/A:1015885204249.

[89] Kissel T, Drewe J, Bantle S, Rummelt A, Beglinger C. Tolerability and Absorption Enhancement of Intranasally Administered Octreotide by Sodium Taurodihydrofusidate in Healthy Subjects. Pharm Res 1992;9:52–7. https://doi.org/10.1023/A:1018927710280.

[90] Dondeti P. Development of a New Non-surgical Perfusion Technique to Evaluate Nasal Drug Delivery. Master's Thesis. University of Rhode Island, 1991.

[91] Hermens WA, Hooymans PM, Verhoef JC, Merkus FW. Effects of absorption enhancers on human nasal tissue ciliary movement in vitro. Pharm Res 1990;7:144–6. https://doi.org/10.1023/a:1015872617511.

[92] Drescher S, van Hoogevest P. The Phospholipid Research Center: Current Research in Phospholipids and Their Use in Drug Delivery. Pharmaceutics 2020;12:1235. https://doi.org/10.3390/pharmaceutics12121235.

[93] Li J, Wang X, Zhang T, Wang C, Huang Z, Luo X, *et al.* A review on phospholipids and their main applications in drug delivery

systems. Asian Journal of Pharmaceutical Sciences 2015;10:81–98. https://doi.org/10.1016/j.ajps.2014.09.004.

[94] Ishii F, Nii T. Properties of various phospholipid mixtures as emulsifiers or dispersing agents in nanoparticle drug carrier preparations. Colloids and Surfaces B: Biointerfaces 2005;41:257–62. https://doi.org/10.1016/j.colsurfb.2004.12.018.

[95] Lee M-K. Liposomes for Enhanced Bioavailability of Water-Insoluble Drugs: In Vivo Evidence and Recent Approaches. Pharmaceutics 2020;12:264. https://doi.org/10.3390/pharmaceutics12030264.

[96] Drejer K, Vaag A, Bech K, Hansen P, Sørensen A r., Mygind N. Intranasal Administration of Insulin With Phospholipid as Absorption Enhancer: Pharmacokinetics in Normal Subjects. Diabetic Medicine 1992;9:335–40. https://doi.org/10.1111/j.1464-5491.1992.tb01792.x.

[97] Alsarra IA, Hamed AY, Alanazi FK. Acyclovir Liposomes for Intranasal Systemic Delivery: Development and Pharmacokinetics Evaluation. Drug Delivery 2008;15:313–21. https://doi.org/10.1080/10717540802035251.

[98] Natsheh H, Touitou E. Phospholipid Vesicles for Dermal/Transdermal and Nasal Administration of Active Molecules: The Effect of Surfactants and Alcohols on the Fluidity of Their Lipid Bilayers and Penetration Enhancement Properties. Molecules 2020;25:2959. https://doi.org/10.3390/molecules25132959.

How to Cite
Velloso, M.I.; Landoni, F. Penetration Enhancers for Development of Intranasal Formulations for Use in Equines. Int J Equine Sci 2022, 1, 16–32.